Functional characterization of *Burkholderia pseudomallei* trimeric autotransporters

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Supplementary Information

Supplemental Table 1. Primers used in this study.

Primer	Sequence (5′-3′)[a]	Description
0796F	CGAAGAGCTCCGTATGCCGGCGTATACGG	Forward and reverse primers to amplify a 420 bp internal fragment of *boaA*
0796R	GCAAGGTACCCCGCGTTGGCCGTGGCCAGC	
1705F	CGAAGGTACCGCCATTACTGGGGATAATG	Forward and reverse primers to amplify a 409 bp internal fragment of *boaB*
1705R	CGAAGAGCTCCAACGCTGCCCGTGCCAGCG	
1434F	CGAAGAGCTCGCGGCGGCATTGGCGGAAC	Forward and reverse primers to amplify a 411 bp internal fragment of *bpaA*
1434R	GCAAGGTACCCGCTGCCCGTCAAATAGCCG	
2063F	CGAAGAGCTCGGCCCGTCACGTACGAGCCC	Forward and reverse primers to amplify a 419 bp internal fragment of *bpaB*
2063R	GCAAGGTACCCGCCTCAGGCCTGCTGCTTG	
1631F	CGAAGAGCTCGCAGGGCATGGTTACGAATAGC	Forward and reverse primers to amplify a 396 bp internal fragment of *bpaC*
1631R	GCAAGGTACCCCCGCGAAATCCACCGTCAG	
0088F	CGAAGAGCTCGGGTTCGCAAAGCACCGCAC	Forward and reverse primers to amplify a 1716 bp internal fragment of *bpaD*
0088R	GCAAGGTACCGACGCATTGGCTCCGAGCGC	
0908F	CGAAGAGCTCCAGCTCGGCCTACGGCGATC	Forward and reverse primers to amplify a 434 bp internal fragment of *bpaE*
0908R	GCAAGGTACCCTTGCGCGCTCGCTTGCGCG	
0909F	TATCATGGTACCTCTATGCACTGATCGGCG	Forward and reverse primers to amplify a 370 bp internal fragment of *bpaEds1*
0909R	AAATAAGAGCTCATGTGCTGGTACTGCTGC	
1439F	CGACGAGCTCGCAGGCGATCAACG	Forward and reverse primers to amplify a 684 bp internal fragment of *bpaF*
1439R	GCAAGGTACCCCAGTGGAAAGCGAGCCGATG	
0908 UP F	AATAATCTCGAGGAGGCTCGGATTATTCCG	Forward and reverse primers to amplify a 502 bp fragment 5′ to *bpaE*
0908 UP R	AACAACCTGCAGGTAGATTTTGTTCACTCG	
0908 DN F	AACAATCTGCAGTATCAGTGGTGATGCGAG	Forward and reverse primers to amplify a 522 bp fragment 3′ to *bpaE*
0908 DN R	ATAATACTCGAGAACTGCTTCACGTCGTCG	
1439 UP F	AATGCCGGATCCATTTAACGTATTCGACGT	Forward and reverse primers to amplify a 394 bp fragment 5′ to *bpaF*
1439 UP R	AACACCCATATGGATCTTGTTCATCTCCGC	
1439 DN F	AACAACCATATGACCTTCGGTGCAGGATAT	Forward and reverse primers to amplify a 403 bp fragment 3′ to *bpaF*
1439 DN R	AATAATGAATTCTGGCCGACGAAGCACACG	
1439ds1/2 UP F	AGGTGGGAATTCAACAGCACGAACAACGTG	Forward and reverse primers to amplify a 584 bp fragment 5′ to *bpaFds1*
1439ds1/2 UP R	CTCGACTCACCGGTAATCGATCACCAGCTA	
1439ds1/2 DN F	TAGCTGGTGATCGATTACCGGTGAGTCGAG	Forward and reverse primers to amplify a 517 bp fragment 3′ to *bpaFds2*
1439ds1/2 DN R	CGGTCCGAATTCACTACTATCCGGACAACG	
1705 UP F	AATAATCCCGGGTATGACGCTGGCTTGTCA	Forward and reverse primers to amplify a 501 bp fragment 5′ to *boaB*
1705 UP R	CCACAACTGCAGCCAGATCACCCGAAATAT	
1705 DN F	CCACAACTGCAGATCTCGTACCAGTGGTAA	Forward and reverse primers to amplify a 505 bp fragment 3′ to *boaB*
1705 DN R	AATAATGAATTCGTGATCCAGCAGCCATGC	

[a]Restriction enzyme recognition sites are underlined if present.

Supplemental Text. Plasmid construction.

Disruption plasmids:

For pCC1, a 420 bp fragment including sequences from codons 126–266 of *boaA* was amplified by PCR from the chromosome of Bp340 using primers 0796F and 0796R. This fragment was digested with SacI and KpnI and ligated into pCC.

For pCC2, a 409 bp fragment including sequences from codons 108–244 of *boaB* was amplified by PCR from the chromosome of Bp340 using primers 1705F and 1705R. This fragment was digested with SacI and KpnI and ligated into pCC.

For pCC3, a 411 bp fragment including sequences from codons 67–204 of *bpaA* was amplified by PCR from the chromosome of Bp340 using primers 1434F and 1434R. This fragment was digested with SacI and KpnI and ligated into pCC.

For pCC4, a 419 bp fragment including sequences from codons 37–176 of *bpaB* was amplified by PCR from the chromosome of Bp340 using primers 2063F and 2063R. This fragment was digested with SacI and KpnI and ligated into pCC.

For pCC5, a 396 bp fragment including sequences from codons 102–234 of *bpaC* was amplified by PCR from the chromosome of Bp340 using primers 1631F and 1631R. This fragment was digested with SacI and KpnI and ligated into pCC.

For pCC6, a 1716 bp fragment including sequences from codons 66–638 of *bpaD* was amplified by PCR from the chromosome of Bp340 using primers 0088F and 0088R. This fragment was digested with SacI and KpnI and ligated into pCC.

For pCC7, a 434 bp fragment including sequences from codons 126–270 of *bpaE* was amplified by PCR from the chromosome of Bp340 using primers 0908F and 0908R. This fragment was digested with SacI and KpnI and ligated into pCC.

For pCC*bpaEds1*, a 370 bp fragment including sequences from codons 67–190 of *bpaEds1* was amplified by PCR from the chromosome of Bp340 using primers 0909F and 0909R. This fragment was digested with SacI and KpnI and ligated into pCC.

For pCC8, a 684 bp fragment including sequences from codons 177–405 of *bpaF* was amplified by PCR from the chromosome of Bp340 using primers 1439F and 1439R. This fragment was digested with SacI and KpnI and ligated into pCC.

Deletion plasmids:

For pCCX3, 502 bp and 522 bp fragments 5′ and 3′ to *bpaE*, respectively, were amplified by PCR from the chromosome of Bp340 using primer pairs 0908 UP F/R and 0908 DN F/R. These fragments were digested with PstI and XhoI and ligated together into pEXKm5, digested with XhoI, in a three-way ligation.

For pMBX1, 394 bp and 403 bp fragments 5′ and 3′ to *bpaF*, respectively, were amplified by PCR from the chromosome of Bp340 using primer pairs 1439 UP F/R and 1439 DN F/R. These fragments were digested with PstI and XhoI and ligated together into pEXKm5, digested with XhoI, in a three-way ligation.

For pMBX2, a 584 bp fragment 5′ to *bpaFds1* and a 517 bp fragment 3′ to *bpaFds2* was amplified by PCR from the chromosome of Bp340 using primer pairs 1439ds1/2 UP F/R and 1439ds1/2 DN F/R, respectively. These fragments were joined by overlap PCR, yielding an approximately 1.1 kb product. This product was digested with EcoRI and ligated into pEXKm5 digested with EcoRI.

For pMBX3, 501 bp and 505 bp fragments 5′ and 3′ to *boaB*, respectively, were amplified by PCR from the chromosome of Bp340 using primer pairs 1705 UP F/R and 1705 DN F/R. These fragments were digested with PstI and XhoI and ligated together into pEXKm5, digested with XhoI, in a three-way ligation.